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Ecology

Parthenogenesis in an outsider crayfish

It has been rumoured<sup>1</sup> that an unidentified decapod crustacean, a crayfish of marbled appearance and of uncertain geographical origin that was introduced into the German aquarium trade in the mid-1990s, is capable of unisexual reproduction (parthenogenesis). Here we confirm that this marbled crayfish (*Marmorkrebs*) is parthenogenetic under laboratory conditions and use morphological and molecular analysis to show that it belongs to the American Cambaridae family. Although parthenogenesis is widespread among the Crustacea<sup>2</sup>, and shrimp, lobsters, crayfish and crabs are otherwise versatile in their modes of reproduction<sup>3–5</sup>, it has not been reported before in decapods, the largest and economically most important

crustacean group. By virtue of its parthenogenetic reproduction, the marbled crayfish emerges not only as an interesting laboratory model but also as a potential ecological threat in that it could outcompete native forms should even a single specimen be released into European lakes and rivers.

To determine the origin and phylogenetic position of the marbled crayfish (Fig. 1a), we compared the sequences of sections of two mitochondrial genes with those of other related species, with particular reference to a similar-looking cambarid species, *Procambarus fallax*, from Florida. The sequences in both genes of the marbled crayfish differ at only a few positions from those of other cambarids, which supports their morphological similarity (the presence of a spermatheca)<sup>6</sup>. Our phylogenetic analysis indicates a particularly close affinity with *P. fallax*, although the marbled crayfish's species identity remains to be verified (Fig. 1b).

We tested whether *Marmorkrebs* could be parthenogenetic by studying a mature female from a laboratory population in Berlin whose spermatheca contained no evidence of spermatophores from copulation, but which repeatedly laid eggs. Between two broods, this crayfish moulted, a procedure that clears any remnants of sperm from the spermatheca. We also sexed laboratory populations in Berlin (93 specimens from 7 mothers of two generations) and Heidelberg (39 specimens), beginning at the earliest stage at which sex can be determined. We found that all specimens (body length, 0.8–8.0 cm) showed female morphology, which excludes protandric hermaphroditism as the mode of reproduction. No spermatophore was detected in our study.

To rule out internal autogamy, we studied the histology and ultrastructure of the reproductive system of all 39 specimens of the Heidelberg population from juvenile (1.9 cm) to post-brooding (6.8 cm) stages. All gonads were normal ovaries with oviducts, and there was no evidence of ovotestes or male gonoducts (results not shown), which are usually present in hermaphroditic crayfish<sup>7</sup>. Azan staining of some adult gonad sections revealed an abundance of large, primary vitellogenic oocytes, and proliferating clusters of small pre-vitellogenic oocytes in the presence of hatchlings, indicating that a new reproductive cycle was already under way (Fig. 1c). These results provide convincing evidence for parthenogenesis in the marbled crayfish.

Our findings have several practical implications. This marbled crayfish will be

useful in the laboratory for physiological, ecological, evolutionary, developmental and genetic studies, having the advantages of fitness, high fertility, fast growth, unisexuality and isogenic progeny<sup>8</sup>. The large oocytes are easily accessible for genetic manipulation, making this species a candidate model for transgenesis<sup>9</sup> in decapod crustaceans. The rapid reproduction of this crayfish might also be of interest for commercial farming purposes.

Last but not least, this crayfish, which is now widespread in Europe's aquaria, could become a menace to European freshwater ecosystems, as the release of even one specimen into the wild would be enough to found a population that might outcompete native crayfish. As an American species, it is a potential transmitter of the infectious crayfish plague that almost caused the extinction of the native European crayfish and which still threatens wild and farmed populations<sup>10</sup>.

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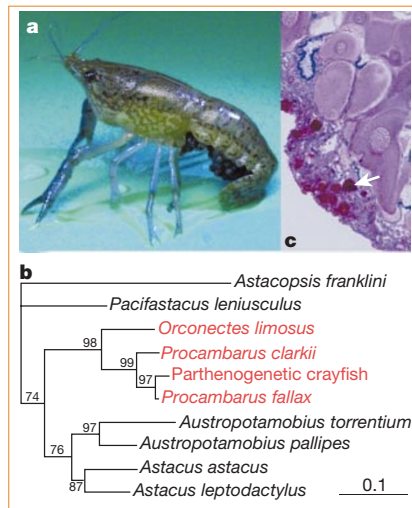
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**Figure 1** Characteristics of the marbled crayfish. **a**, Lateral view of the crayfish, showing the eggs attached to its pleopods. Adult specimens are 3–10 cm in length (rostrum to telson). The basic colour is brown–green, with light and dark spots. **b**, Phylogenetic relationships of the marbled crayfish based on a comparison of the partial sequences of two mitochondrial genes, *Cox1* and the gene encoding 12S ribosomal RNA; GenBank accession numbers, AY151515–AY151524, AY151525–AY151534. Maximum-likelihood analysis used the Hasegawa–Kishino–Yano model, assuming rate heterogeneity with a log likelihood of –3075.69; the reliability of branches is given as a percentage of puzzling steps where the appropriate node arises. Parsimony analysis produced similar results. The marbled crayfish belongs to the American Cambaridae (red) and is closely related to *Procambarus fallax* (only 2.2% of base positions different). Scale bar, 0.1 nucleotide substitutions. **c**, Azan-stained histological section of a gonad of a female with hatchlings exclusively composed of ovarian tissue. Arrow, pre-vitellogenic oocyte; arrowhead, primary vitellogenic oocyte.

correction

Ultrahard polycrystalline diamond from graphite

T. Irifune, A. Kurio, S. Sakamoto, T. Inoue, H. Sumiya *Nature* 421, 599–600 (2003)

In the legend to Fig. 1a of this communication, the diameter of the transparent polycrystalline diamond shown is 1 mm, and not 0.1 mm as published; the scale divisions represent 0.1 mm. Also, the first full paragraph in the second column on page 600 should read: "Recent chemical-vapour deposition techniques provided pure polycrystalline diamonds, but these diamonds are not sintered and have poor intergrain adhesion. Accordingly, they have been reported to have a hardness of ~80–100 GPa (ref. 10), which is significantly lower than the highest value (~120 GPa; ref. 4) for single-crystal diamonds."